

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

Claim 1. (currently amended) A composition for detecting β -1,3-glucan exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions, prepared by the method comprising:

(i) collecting a sample comprising a mixture of plasma and hemocyte lysate from an insect;

(ii) treating said sample with a solvent or buffer solution containing a sufficient amount of a chelating agent to chelate calcium ions existing in said sample and during a present separation process to obtain fractions therefrom; and

(iii) selecting fractions exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions from the obtained fractions.

Claim 2. (original) A composition for detecting β -1,3-glucan the minimum down to 20 pg/ml in the presence of calcium ions.

Claim 3. (original) A method of preparing a phenoloxidase composition activated by β -1,3-glucan in the presence of calcium ions, comprising: collecting a sample comprising a mixture of plasma and hemocyte lysate from an insect; treating said sample with a solvent or buffer solution containing a sufficient amount of a chelating agent to chelate

calcium ions existing in said sample and during a separation process to obtain fractions therefrom; and selecting fractions exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions from the obtained fractions.

Claim 4. (original) The method according to claim 3 wherein said insect belongs to Coleoptera.

Claim 5. (original) The method according to claim 4 wherein said Coleoptera belongs to Tenebrionidae or Scarabaeidae.

A (Claim 6. (original) The method according to claim 3 wherein said fractions are obtained by column chromatography.

Claim 7. (original) The method according to claim 6 wherein the column used for said column chromatography is packed with a resin comprising dextran or vinyl.

Claim 8. (original) The method according to claim 3, wherein whole or partially purified hemocyte lysate is further added to the fractions exhibiting the phenoloxidase activity by β -1,3-glucan in the presence of calcium ions.

Claim 9. (original) A method of preparing a phenoloxidase composition

activated by β -1,3-glucan in the presence of calcium ions, comprising: treating insect plasma with a solvent or buffer solution containing a sufficient amount of a chelating agent to chelate calcium ions existing in said plasma and during a separation process to obtain fractions therefrom; adding hemocyte lysate or partially purified hemocyte lysate to said fractions; and selecting fractions exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions.

Claim 10. (original) The method according to claim 9 wherein said insect belongs to Coleoptera.

A (Claim 11. (original) The method according to claim 10 wherein said Coleoptera belongs to Tenebrionidae or Scarabaeidae.

Claim 12. (original) The method according to claim 9 wherein the said fractions are obtained by column chromatography.

Claim 13. (original) The method according to claim 12 wherein the column used for said column chromatography is packed with a resin comprising dextran or vinyl.

Claim 14. (original) The method according to claim 9, wherein whole or partially purified hemocyte lysate is further added to the fractions exhibiting the phenoloxidase activity

by β -1,3-glucan in the presence of calcium ions.

Claim 15. (original) A method of detecting β -1,3-glucan comprising the steps of: collecting a sample from a specimen; adding a composition of claim 1 and calcium ions to said sample; and measuring the phenoloxidase activity in the sample.

Claim 16. (original) A diagnostic kit for detecting β -1,3-glucan which contains the composition detecting β -1,3-glucan the minimum down to 20 pg/ml in the presence of calcium ions.

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Claim 17. (original) A diagnostic kit for detecting β -1,3-glucan exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions, prepared by the method comprising: collecting a sample comprising a mixture of plasma and hemocyte lysate from an insect; treating said sample with a solvent or buffer solution containing a sufficient amount of a chelating agent to chelate calcium ions existing in said sample and during a separation process to obtain fractions therefrom; and selecting fractions exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions from the obtained fractions.

Claim 18. (original) The diagnostic kit according to claim 17 wherein said insect belongs to Coleoptera.

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Claim 19. (original) A diagnostic kit for detecting β -1,3-glucan exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions, prepared by the method comprising: treating insect plasma with a solvent or buffer solution containing a sufficient amount of a chelating agent to chelate calcium ions existing in the plasma and during a separation process to obtain fractions; adding hemocyte lysate or partially purified hemocyte lysate to the above obtained fractions; and selecting fractions exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions.

Claim 20. (original) The diagnostic kit according to claim 19 wherein the insect belongs to Coleoptera.

Claim 21. (original) A composition for detecting β -1,3-glucan exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions, prepared by the method comprising: treating insect plasma with a solvent or buffer solution containing a sufficient amount of a chelating agent to chelate calcium ions existing in the plasma and during a separation process to obtain fractions therefrom; adding hemocyte lysate or partially purified hemocyte lysate to said fractions; and selecting fractions exhibiting phenoloxidase activity by β -1,3-glucan in the presence of calcium ions.

Claim 22. (original) A method of detecting β -1,3-glucan comprising the steps of: collecting a sample from a specimen; adding a composition of claim 21 and calcium ions

to said sample; and measuring the phenoloxidase activity in the sample.



Claim 23. (original) The composition according to claim 2 wherein said composition is characterized in measuring the phenoloxidase activity.

Claim 24. (original) The diagnostic kit according to claim 16 wherein said diagnostic kit is characterized in measuring the phenoloxidase activity.
